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SUMMARY OF DATA ON POTASSIUM SORBATE

Abstract. Potassium sorbate, a widely used preservative in foods, is used in the tobacco industry as a preservative to extend the shelf life of tobacco. Commercial cigarettes to which potassium sorbate has been added may contain 0.23 mg of potassium sorbate. This is much less than 0.01% of the cigarette by weight, and is also far lower than the amount of potassium sorbate consumers are exposed to in foods. Potassium sorbate is approved for cigarette use in Great Britain, at levels up to 0.5%, and in Germany.

Research comparing cigarette smoke condensate from reference cigarettes with condensate from cigarettes containing 1.5, 3, or 6 times the amount of potassium sorbate used in a typical commercial cigarette has indicated that the addition of potassium sorbate does not affect the biological activity of the condensate. Studies also suggest that the levels of potassium sorbate used in cigarettes present no risk to humans. Potassium sorbate has virtually no toxic effects except at levels thousands of times larger than smokers encounter, and studies have not shown potassium sorbate to be carcinogenic, mutagenic, or teratogenic.

Background. Potassium sorbate ($CH_3CH=CHCH=CHOOK$; CAS No. 590-00-1), also known as potassium trans, trans-2,4-hexadienoate, is a white fluffy powder, highly soluble in water (58%) and slightly soluble in ethanol (6%). The melting point is 270°C with decomposition (Scientific Literature Review, 1973). Potassium sorbate is both tasteless and odorless (Chichester, 1972).

Potassium sorbate has a long history of use as an antimicrobial food additive. Sorbic acid and its salts have broad-spectrum activity against yeasts and molds, but are less

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active against bacteria (Chichester, 1972). The antimicrobial activity of sorbates has been demonstrated below pH 6.5 and increases with the concentration of the undissociated acid as the pH of the medium is lowered (SCOGS, 1975).

Sorbic acid was isolated in 1859 from the berries of the mountain ash. It occurs there naturally as the lactone, parasorbic acid (SCOGS, 1975). There is no evidence that it occurs naturally either in tobacco or in mammalian tissues.

Sorbic acid is prepared commercially by a number of synthetic procedures including: (a) the condensation of crotonaldehyde and ketene in the presence of boron trifluoride (Hagemeyer, 1984); (b) the condensation of crotonaldehyde and ketene in the presence of zinc isovalerate (Fernholz, 1959); and (c) the manganese catalyzed oxidation of 1,1,3,5-tetra-ethoxyhexane followed by the base catalyzed elimination of ethanol from the resulting 3,5-dimethoxyhexanoic acid (Parker 1960). Potassium sorbate is prepared by the neutralization of sorbic acid with potassium hydroxide.

Potassium sorbate is used as a preservative in a wide variety of foods, including cake mixes, carbonated beverages, fruit juices, wines, salads and fruit cocktails, dried fruits, margarine, canned fish products, jams and jellies, puddings, soft candy, and snack foods (Chichester, 1972; SCOGS, 1975). It has recently been proposed that potassium sorbate be approved for use in bacon, processed

poultry, frankfurters and sausage (Hartman, 1983). The Select Committee on GRAS Substances (SCOGS, 1975) estimated the average daily intake of sorbic acid and potassium sorbate to be 25 mg. The Joint FAO/WHO Expert Committee (1974) on food additives has estimated that the acceptable daily intake for sorbic acid and its salts, including potassium sorbate, is 25 mg/kg body weight.

Tobacco Uses. Potassium sorbate is used in the tobacco industry as a preservative to extend the shelf life of tobacco. Approximately 250,000 pounds of potassium sorbate were used in the industry in 1986. A representative level for potassium sorbate in tobacco is about 0.3 pounds per thousand pounds of tobacco. Therefore, some commercial cigarettes may contain 0.23 mg potassium sorbate per cigarette, or much less than 0.01% of the cigarette by weight.

Regulatory Status. FDA has classified potassium sorbate as Generally Recognized as Safe (GRAS) for use both as a chemical preservative in foods and in food packaging materials. 21 C.F.R. §§ 182.3640 and 182.90. In addition, potassium sorbate is on the FEMA list of GRAS substances. As of 1972, seventeen countries in addition to the United States had approved sorbates for use as antimicrobial food additives, including Canada, the United Kingdom, West Germany and Japan (Chichester, 1972). Potassium sorbate is approved for use in cigarettes by the U.K.'s Independent Scientific Committee on Smoking and Health, up to a level of 0.5%, and under the German Tobacco Ordinance.

Acute and Subchronic Animal Studies. The oral LD_{50} of potassium sorbate in rats for the trans, trans-isomer was (3) 4.9 g/kg. The intraperitoneal LD_{50} value in mice was 1.3 g/kg (Scientific Literature Review, 1973). Rats administered a single dose of 200 mg of potassium sorbate had slightly elevated bilirubin, cholesterol, pancreatic juice, pancreatic protein, lipase and amylase, and decreased chymotrypsin activity (Faur, 1966).

Potassium sorbate was fed to groups of 10 rats (5 male and 5 female) at levels of 0, 1, 5, and 10% of the diet for three months. Relative liver weights were the same in all groups. Kidney weights were increased at the 10% level and to a lesser degree at the 5% level. Weight gains of female animals were depressed initially when fed at the 10% level and to a lesser degree at the 5% level. In the same report eight dogs received 1% and eight dogs 2% potassium sorbate in the diet for three months. The test animals gained the same weight as four control dogs. At autopsy, gross examination revealed no deleterious effects attributable to potassium sorbate (SCOGS, 1975).

Another three month feeding study was carried out using levels of 0.25% and 2% potassium sorbate in rats. At the 2% level decreases in pancreatic protein chymotrypsin, amylase and lipase were observed, while slight increases in bilirubin and cholesterol were noted. The only change noted at the 0.25% level was an increase in pancreatic juice, its protein content and its enzymes (Faur, 1966).

The primary metabolic products of sorbate in animals have been shown to be carbon dioxide and water. A small amount of trans, trans-muconic acid (0.1 - 0.2%) has been verified as a metabolic product in rabbits. The metabolism of sorbates is identical in animals and human beings (Scientific Literature Review, 1973).

Chronic Exposure Studies/Carcinogenicity. Most long term studies have focused on sorbic acid rather than the salts of sorbic acid. In a 17-month study, 25 male and 25 female mice were fed sorbic acid (40 mg/kg body weight) daily as a paste prior to the main feed (Shtenberg, 1970). Compared to controls the weights of liver, kidney and testes relative to body weight were lower for the experimental animals sacrificed at the end of the test. These changes were not considered pathological by the investigators.

One hundred rats maintained on 0.1, 0.5 and 5.0 percent sorbic acid in the diet (50, 250 and 2500 mg/kg body weight per day) for a test period of 1000 days did not differ from controls in growth, reproduction, health, life expectancy or cause of death (Lang, 1960). A group of rats maintained through the second generation on the 0.1 or 0.5 percent diets exhibited no differences from controls in growth or reproduction. Thirty rats of the second generation received 5 percent sorbic acid in the diet for 252 days with no demonstrable pathological findings. An unpublished report from the same laboratory concerned 100 rats (50 males, 50 females) fed a

diet providing 5 percent sorbic acid during their life span. The average life span of males compared to controls was 811 vs. 709 days and for females 789 vs. 804 days. There were no differences in organ weights of individual groups. In each group (5 percent sorbic acid and controls) only two tumors were found. There were no abnormalities in liver, kidney, heart or testes (SCOGS, 1975).

Dickens (1966) attempted to study the carcinogenicity of sorbic acid; however, the results were inconclusive. In a repetition of these test using sorbic acid from another source, no tumors were obtained in Wistar rats when sorbic acid (2 mg in 0.5 ml in peanut oil) was injected subcutaneously twice weekly for 56 to 60 weeks (Dickens, 1968). In the same study potassium sorbate did not induce tumors when fed for 60 weeks to rats at 0.1 percent of the diet or when dissolved in their drinking water at 0.3 percent.

In another carcinogenicity study, mice that had been fed a control diet or a diet supplying up to 300 mg/kg sorbic acid for 3 months received ascitic Ehrlich cancer (4) intraperitoneally by transplant (Dinerman, 1966). Within a 66-day observation period, the percentage of animals that developed tumors in control and experimental groups did not differ.

No carcinogenic effect was demonstrated in rats given dietary sorbic acid levels up to 10 percent (about 5 g/kg body weight daily) for 2 years (Gaunt, 1975). Groups of 48 male (90 to 145 g) and 48 female rats (80 to 130 g) were

fed diets containing 0, 1.5 or 10 percent sorbic acid. At the 10 percent level the thyroid weight in males, the relative liver weights in both sexes, and the relative kidney, small intestine, and ovary weights in females were increased slightly. No significant differences were found between the control and treated groups by hematological examinations, analyses of serum, studies of renal function by histopathological examination, or in mortality rate.

(4a) Mutagenicity. A number of studies have shown potassium sorbate to be non-mutagenic in a variety of bacterial test systems (Hartman, 1983). However, other work has indicated a low level of genotoxic activity for potassium sorbate in several test systems. (5)

A 1977 report demonstrated that potassium sorbate caused chromosomal aberrations in pseudo-diploid (6) (7) Chinese hamster cells, albeit only at the highest concentration (0.02 M). No sister chromatid exchanges (SCE) were observed at any concentration. The authors point out that the level at which chromosomal aberrations were noted was much higher than the amount man is exposed to on a daily basis (Abe, 1977).

A study of the potential genotoxicity of potassium sorbate as measured by chromosome aberrations and SCE was carried out by Hasegawa (1984). This study used V79 Chinese hamster cells. As in the Abe study (1977), potassium sorbate was found to be clastogenic. However, chromosome aberrations were caused only at the highest dose (20 mg/ml), and about 50%

of the clastogenic effect can be attributed to an increase in osmotic pressure. On the other hand, unlike the Abe study, potassium sorbate was found to cause a small but significant increase in SCE at a dose of 10 mg/ml. There was no dose response effect for the increase in SCE over the dose range 10 mg/ml to 20 mg/ml. This study also demonstrated that potassium sorbate was not mutagenic to V79 cells.

Another report describes a study concerning the mutagenic effects of the intestinal contents of mice fed both potassium sorbate and sorbic acid. Mice were fed a diet containing 1.34%, 6.70% and 20.1% potassium sorbate for six months. The intestinal contents were removed from 5 to 10 mice from each group at one week and 1, 3 and 6 months, and ether extracts were tested for mutagenicity by the Ames assay using TA98 and TA100 with and without S9. No activity was detected in any of these samples (Tsuchiya, 1983). A group of animals fed a diet consisting of 15% sorbic acid was tested similarly. Although the ether extracts were also negative in the Ames assay, the acid fraction from the ether extracts was weakly but statistically significantly positive. Earlier results had shown that both potassium sorbate and sorbic acid were not mutagenic when tested in their pure form in the Ames assay using TA98 and TA100 with and without S9 (Kawachi, 1980).

In 1984 Tsuchiya and Yamaha published a follow-up to their 1983 study. In this case feeding was carried out for

12 months as compared to six months, and the acidic fraction of the ether extracts from all test groups was evaluated in the Ames assay. No mutagenicity of the acidic fractions of the ether extract of the intestinal contents of mice fed 1.34% and 6.70% potassium sorbate was detected. At the 20% potassium sorbate level, a slight mutagenic effect was observed, but only at the highest dose (8.0 mg/plate). However, there was a dose related depletion of liver glutathione levels for all doses tested.

One finding from the testing of potassium sorbate is that mutagenic substances can be formed by the interaction of sorbate and nitrite at pH's which mimic gastric conditions. Two compounds which have been shown to be mutagenic, ethyl-nitrolic acid and 1,4-dinitro-2-methylpyrrole, have been isolated from this interaction. Formation of these compounds is blocked by ascorbate at low pH (Hartman, 1983). On the other hand a recent report on the mutagenicity of sodium nitrite and potassium sorbate in combination indicates that mutagen formation may not occur under physiological conditions (Budayova, 1985). Sodium nitrite was found to be cytotoxic to V79 cells, and a synergistic effect on cytoxicity was observed for sodium nitrite and potassium sorbate in combination. However, despite the fact that sodium nitrite alone was slightly mutagenic to V79 cells, the combination of sodium nitrite and potassium sorbate was inactive.

Reproduction and Teratology. Studies of potassium sorbate for teratogenicity by chick embryo tests were negative. Daily administration of up to 460 mg/kg body weight to pregnant mice (albino CD-1) or up to 340 mg/kg body weight to pregnant rats (Wistar derived stock) for 10 consecutive days (days 6 to 15 of gestation) had no clearly discernable effect on nidation or on maternal or fetal survival. The number of abnormalities seen in either soft or skeletal tissues of the test group did not differ from the number occurring spontaneously in sham-treated controls (SCOGS, 1975).

Pyrolysis Chemistry Data. Potassium sorbate was subjected to Curie Point Pyrolysis/GG/MS at temperatures of 315°C and 590°C. The resulting chromatograms contained no peaks, indicating that potassium sorbate does not decompose into any volatile products during pyrolysis.

Pyrolysis Toxicology Data. The effect of adding potassium sorbate to cigarettes was evaluated by testing the resulting cigarette smoke condensate (CSC) in the Ames assay. Potassium sorbate was dissolved in water and injected into University of Kentucky 1R4F Research Cigarettes at levels of 1.5, 3.0 and 6.0 times those normally used in commercial cigarettes. The cigarettes were smoked, and the impaction-trapped CSC was tested in the Ames assay (strains TA98 and TA100) with and without metabolic activation. The results indicated that the addition of potassium sorbate to cigarettes, at any of the levels tested, did not alter the activity of the CSC.

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